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(54) Title: PEPTIDES INHIBITING IL-1 BETA RELEASE

(57) Abstract

Di-, tri- and tetrapeptides in which the last α-amino acid is based on aspartic acid and attached to a residue A_5 which is H; CF3; $-Z_1-Z_2-Y_2$ wherein each of Z_1 and Z_2 independently is a direct bond or an α-amino acid residue and Y_2 is NH_2, C_{1-4} alkylamino, di-(C_{1-4} alkyl)amino or a heterocyclic radical attached by a nitrogen to Z_2; $-CH_2-X_1-Y_3$ wherein X_1 is O or S and Y_3 is heteroaryl; $-CH_2-Y_3$; substituted phenyl; ring substituted phenoxymethylene or phenylthiomethylene; ring substituted pyridyloxymethylene; or a radical $-CH_2-X_1-CO-Y_4$ wherein X_1 is O or S and Y_4 is trialkylmethyl or substituted phenyl or pyridyl, in free form or in salt form, have pharmacological activity, e.g. IL-1β release inhibiting properties.

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PEPTIDES INHIBITING IL-1 BETA RELEASE

The present invention relates to peptides having pharmaceutical utility, processes for their production, pharmaceutical compositions comprising them and their use as pharmaceuticals.

More particularly the present invention provides a compound of formula I

$$R-[A_1-A_2]_n-A_3-A_4-X-A_5 \qquad (I)$$

wherein

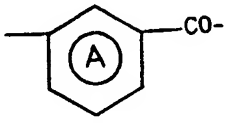
R is hydrogen, an amino protecting group or optionally ring substituted benzyloxy

A_1 is Val, Leu, Ala, Ile or trimethylsilyl-Ala

A_2 is Phe or Tyr,

n is 0 or 1,

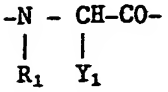
A_3 is a direct bond, Val, Leu, Ala, Ile, trimethylsilyl-Ala or a divalent radical of formula (a)



(a)

wherein ring A is optionally substituted by hydroxy or C_{1-4}alkoxy,

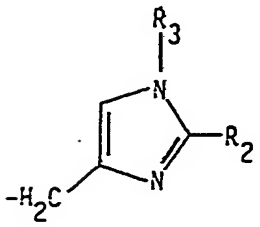
A_4 is a direct bond or a divalent radical of formula (b)



(b)

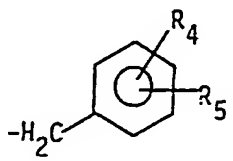
wherein R_1 is hydrogen or C_{1-4}alkyl, and

Y_1 is the residue attaching to the α-carbon atom of an α-amino acid and optionally protected, $-CH_2-CH_2-N(C_{1-4}alkyl)_2$, imidazol-2-yl-methyl, benzimidazol-2-yl-methyl, 1H-1,2,4-triazol-3-yl-methyl, pyrazol-3-yl-methyl, indazol-3-yl-methyl or a radical of formula (c) or (d)



(c)

or



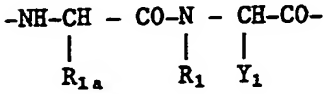
(d).

wherein

each of R_2 and R_3, independently, is hydrogen, halogen, C_{1-4}alkyl, CF_3 or trityl, at most one of R_2 and R_3 being H, and

each of R_4 and R_5 independently is hydrogen, C_{1-4}alkyl, hydroxy, C_{1-4}alkoxy, CF_3, phenyl or halogen, at most one of R_4 and R_5 being H,

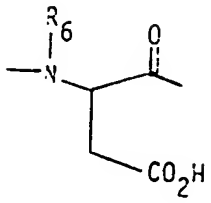
or A_3 and A_4 form together a radical of formula (aa)



(aa)

wherein Y_1 is as defined above and R_1 and R_{1a} form together $-(CH_2)_m-$ wherein m is 2, 3, 4 or 5, and

1) X is a divalent radical of formula (e_1)

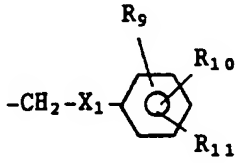


(e_1)

wherein R_6 is H or C_{1-4}alkyl,

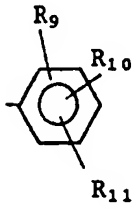
and A_5 is hydrogen; CF_3; a radical $-Z_1-Z_2-Y_2$ wherein each of Z_1 and Z_2 independently is a direct bond or an α-amino acid residue and Y_2 is NH_2, C_{1-4}alkylamino, di-(C_{1-4}alkyl)amino or a heterocyclic radical attached by a nitrogen to Z_2; a radical $-CH_2-X_1-Y_3$ wherein X_1 is O or S and Y_3 is heteroaryl; a radical $-CH_2-Y_3$; or a radical of formulae (k) to (m)

(k)

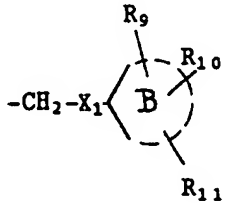


(l)

or



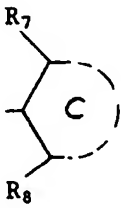
(m)



(o)

wherein

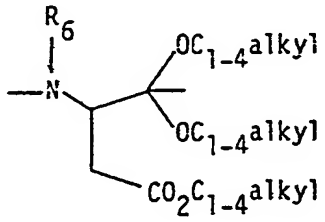
Y_4 is tri-(C_{1-4}alkyl)methyl or a residue



ring B is pyridyl,

ring C is phenyl or pyridyl,
each of R_7 and R_8 independently is C_{1-4}alkyl, C_{1-4}alkoxy, CF_3, halogen, nitro or cyano, and
each of R_9, R_{10} and R_{11} independently is nitro, cyano, CF_3, carbamoyl, CO_2R_{12}, -CH=CH-CN or $-CH=CHCO_2R_{12}$ wherein R_{12} is C_{1-6}alkyl,

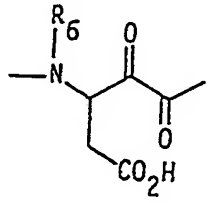
X being also a divalent radical of formula (e_2)



(e_2)

when A_5 is H, or

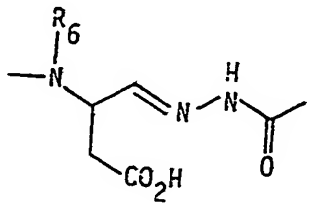
2) X is a divalent radical of formula (f),



(f)

and A_5 is $-Z_1-Z_2-Y_2$ or a radical of formulae (k) to (o) as defined above, or OR_{13} or $NR_{14}R_{15}$ wherein R_{13} is C_{1-12}alkyl optionally substituted by OH or interrupted by O and each of R_{14} and R_{15} is independently hydrogen, C_{1-12}alkyl, C_{5-7}cycloalkyl or benzyl, or

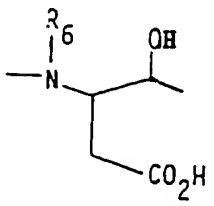
3) X is a divalent radical of formula (g)



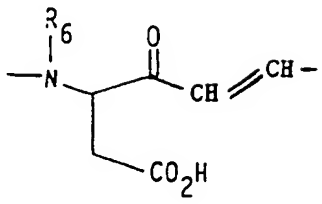
(g)

and A_5 is $-Z_1-Z_2-Y_2$ as defined above, or

4) X is a divalent radical of formula (h) or (j)



(h)



(j)

and A_5 is a radical of formulae (k) to (o), $-CH_2-Y_3$ or $-CH_2-X_1-Y_3$ as defined above,

with the provisos that
only one of A_3 and A_4 can be a direct bond when n is 0, and
each of A_3 and A_4 is other than a direct bond when n is 1.

and the physiologically-hydrolysable and -acceptable esters or amides thereof,

in free form, in salt form or in the form of complexes.

Examples of protecting groups as R are e.g. disclosed in "Protective Groups in Organic Synthesis", T. W. Greene, J.Wiley & Sons NY (1981), 219-287, for example acyl such as acetyl, methoxysuccinyl, hydroxysuccinyl or benzoyl optionally substituted on the phenyl ring with e.g. p-methoxycarbonyl, p-methoxy or p-nitro; alkoxycarbonyl such as t-butyloxycarbonyl; arylmethoxycarbonyl such as 9-fluorenylmethoxycarbonyl or benzyloxy carbonyl optionally substituted on the phenyl ring with p-methoxy, p-nitro, p-chloro or m-phenyl; arylmethyl such as benzyl optionally ring substituted with p-methoxy, p-nitro or p-chloro; or arylsulfonyl such as phenylsulfonyl optionally ring substituted

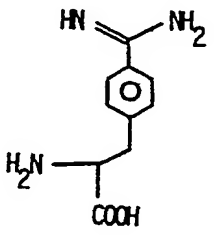
with p-methyl or p-methoxy, or naphthylsulfonyl optionally ring substituted with e.g. amino or di(C_{1-4}alkyl)amino.

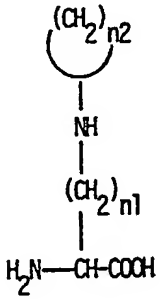
When R is ring substituted benzyloxy, it is preferably benzyloxy substituted with hydroxy or C_{1-4}alkoxy. Preferably R is unsubstituted benzyloxy.

Halogen is preferably fluorine or chlorine.

When A_3 is a substituted radical of formula (a) it is preferably substituted by C_{1-4}alkoxy, preferably in para to -C=O.

By α-amino acid is meant a naturally occurring or commercially available or non natural α-amino acid or an optical isomer thereof. A non natural α-amino acid is an α-amino acid which is not incorporated into a protein under mRNA direction, e.g. β-Nal, a fluoro-α-amino acid such as fluoroalanine, trimethylsilyl-Ala or an α-amino acid such as





wherein n_1 is an integer from 1 to 6 and n_2 is an integer from 1 to 12.

Protecting groups which may be present in Y_1 are groups which protect the O, S or N functionality in the side chain amino groups of an α-amino acid. N-protecting groups are e.g. as disclosed above for R, or C_{3-5}alkyl such as isopropyl, formyl, a sugar residue such as 1-deoxy-fructosyl or α-glucosyl(1-4)-

deoxyfructosyl, dihydroxy-C_{3-6}alkyl such as dihydroxypropyl, C_{5-7}cycloalkyl such as cyclohexyl or tropinyl. O- and S-protecting groups for hydroxy and thiol functionalities are known and may be e.g. methyl, t.-butyl or benzyl.

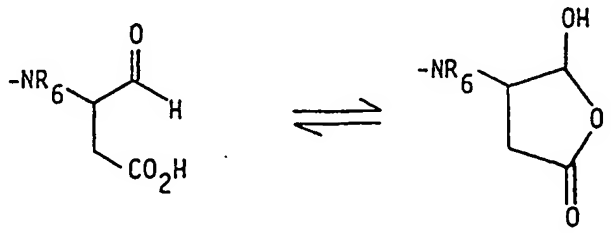
When Y_2 is a heterocyclic radical, it may be e.g. a 5 or 6 membered ring, e.g. piperidino or pyrrolidinyl.

Examples of heteroaryl as Y_3 include e.g. 5-, 6- or 7-membered unsaturated heterocyclic radicals, comprising at least one nitrogen and optionally further heteroatoms such as N, O or S. Preferably Y_3 is heteroaryl comprising from 1 to 4 nitrogen atoms, e.g. pyridyl, triazolyl, tetrazolyl, triazin-dionyl.

In ring B of radical (o), the nitrogen atom may be in o-, m- or para. When ring C in radical (k) is pyridyl, it may be 3-, 4- or 5-pyridyl.

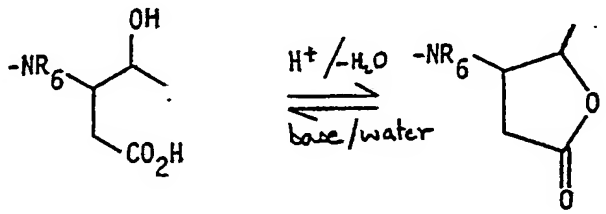
Radicals (e_1), (e_2), (f), (g) and (j) are derived from Asp and comprise one asymetric carbon atom and radical (h) comprises two asymetric carbon atoms and accordingly they lead to optical isomerism. It will be understood that the present invention includes all individual isomeric forms and diastereoisomers as well as mixtures, e.g. racemates, unless otherwise stated.

Radical of formula (e_1) attached to A_5 which is hydrogen may exist in both cyclic as well as in non-cyclic form e.g. as follows:



It is to be understood that where tautomeric forms occur, the present invention embraces both lactol and oxo-carboxylic acid forms, i.e. although compounds of formula I wherein X is a radical of formula (e_1) are defined for convenience by reference to the oxo-carboxylic acid form only, the invention is not to be understood as being in any way limited by the particular nomenclature or graphic representation employed. Similar considerations apply in relation to starting materials exhibiting lactol/oxo-carboxylic acid tautomerism as hereinafter described.

The same considerations also apply to radical of formula (h) which may exist in both linear and cyclic form as follows:



and to compounds of formula I comprising a radical of formula (h) and to the corresponding starting materials.

By the term "physiologically-hydrolysable and -acceptable esters or amides" are meant esters and amides which are hydrolysable under physiological conditions to yield alcohols or amines which are themselves physiologically acceptable, i.e. which are non-toxic at the desired dosage levels.

Such esters or amides are obtained by esterification or amidation, respectively, of a compound of formula I wherein X is a radical bearing a carboxy group. Such esters include esters with an aliphatic or alicyclic alcohol or polyol having 1 to 12 carbon atoms. Such amides include amides with aliphatic amines, e.g. C_{1-4}alkyl amine, C_{1-4}alkoxy-C_{1-4}alkyl amine such as

β-methoxy-ethyl amine, or aniline.

The compounds of formula I may exist e.g. in free form, acid addition salt form or in the form of complexes thereof. Acid addition salts may be formed with e.g. organic acids, polymeric acids and inorganic acids. Such acid addition salt forms include e.g. the hydrochlorides and acetates. Salt forms may also include those obtainable with the carboxylic group present in compounds of formula I, e.g. alkali metal salts such as sodium or potassium, or substituted or unsubstituted ammonium salts. Complexes are e.g. formed from compounds of formula I on addition of inorganic substances, e.g. inorganic salts or hydroxides such as Ca- and Zn-salts, and/or an addition of polymeric organic substances.

In the compounds of formula I, the following significances are preferred either individually or in any combination or subcombination:

1. R is an amino protecting group or benzyloxy, preferably an amino protecting group. R is preferably benzyloxy when n is 0 and A_3 is a radical of formula (a).

2. Each of A_3 and A_4 are other than a direct bond.
3. n is 0.
4. A_3 is a radical of formula (a) when n is 0.

5. A_3 is a direct bond, Val, Leu, Ala, Ile or trimethylsily-Ala.

6. R_1 is hydrogen or methyl, preferably hydrogen.

7. Y_1 is the residue attaching to the α-carbon atom of an α-amino acid selected from Ala, Leu, His, Phe, Met, Trp, trimethylsilyl-Ala and optionally side chain protected Arg, Orn and Lys, or Y_1 is a radical of formula (c).

8. A_3 and A_4 may also form together a radical of formula (aa) when n is 0.

9. m is 2 or 3.

10. X is a radical of formula (e_1) and A_5 is other than H.

11. X is a radical of formula (g).

12. X is a radical of formula (h) or (j).

13. R_6 is hydrogen or methyl, preferably hydrogen.

14. Z_1 in A_5 is the residue of a natural α-amino acid, preferably of an aromatic/heterocyclic α-amino acid, particularly Pro.

15. Z_2 in A_5 is the residue of a natural α-amino acid, preferably an aliphatic α-amino acid, particularly an aliphatic α-amino acid without further functional group, most preferably Val.

16. X is a radical of formula (e_1) optionally esterified or amidated and A_5 is a radical of formula (k), (l) or (o).

17. X is a radical of formula (e_1), (h) or (j) optionally esterified or amidated and A_5 is $-CH_2-X_1-Y_3$, preferably $-CH_2-S-Y_3$, or $-CH_2-Y_3$.

18. X is a radical of formula (h) or (j) optionally esterified or amidated and A_5 is (k).

19. X is a radical of formula (j) optionally esterified or amidated and A_5 is a radical of formula (m).

20. Radical of formula (m) is monosubstituted, preferably in

para, more preferably it is 4-nitrophenyl.

21. All α-amino acid residues present except X have the L configuration. X has the D or L configuration.

22. The asymetric carbon in (h) bearing the OH group has the configuration S. Preferably the asymetric carbon in (j) bearing R_6 has the configuration R.

In a series of specific embodiments, the present invention also provides a compound of formula I wherein
n is 0, A_3 is a direct bond, Val, Leu, Ala, Ile or trimethylsilyl-Ala, A_4 is as defined above, or A_3 and A_4 form together a radical of formula (aa) as defined above, and

i) X is a radical of formula (e_1) or (e_2) as defined above and A_5 is H, or
ii) X is a radical of formula (e_1) or (f) and A_5 is $-Z_1-Z_2-Y_2$ as defined above or a radical of formula (k), (l) or (m) wherein X_1 is O,
iii) X is a radical of formula (g) and A_5 is $-Z_1-Z_2-Y_2$, or
iv) X is a radical of formula (j) and A_5 is a radical of formula (k), (l) or (m) wherein X_1 is O.

The present invention also provides a process for the production of a compound of formula I, which process comprises:

a) removing at least one protecting group from a compound of formula I in protected form or adding a protecting group R at the N-terminal group of a compound of formula I; or

b) converting one compound of formula I into another compound of formula I; or

c) coupling together by an amide bond two peptide fragments,

each of which contains at least one amino acid in protected or unprotected form and one peptide fragment containing a radical of formula (e_1) to (j) as defined above, the peptide fragments being such that a protected or unprotected peptide having the sequence according to formula I above is obtained and, if necessary, removing the protecting group or groups from a compound of formula I in protected form; or

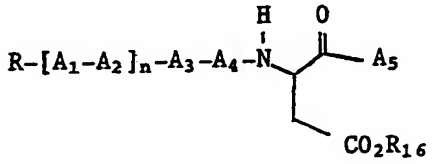
d) for the production of a compound of formula I wherein X is a radical of formula (e_1) or (h) and A_5 is a radical of formula (k), (l) or (o) or $-CH_2-X_1-Y_3$, reacting a compound of formula III

$$R-[A_1-A_2]_n-A_3-A_4-X'-CH_2-Z_a \qquad (III)$$

wherein R, A_1 to A_4 and n are as defined above, X' is a radical of formula (e_1) or (h), and Z_a is a leaving group, e.g. halogen,

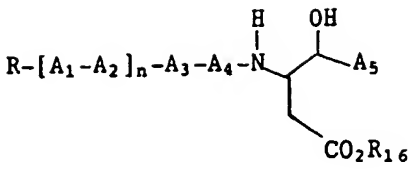
with a corresponding phenol, thiophenol or HX_1-pyridine or an acid of formula HX_1-CO-Y_4 or a functional derivative thereof or HX_1-Y_3; or

e) for the production of a compound of formula I



(I)

wherein R, A_1 to A_5 and n are as defined above and R_{16} is a C_{1-12} aliphatic or alicyclic residue, oxidizing a compound of formula V



(V)

wherein R, A_1 to A_5, n and R_{16} are as defined above,

and recovering a compound of formula I thus obtained in free or salt form or in the form of a complex.

Processes (a) to (e) above may be carried out in accordance with standard techniques known in the art.

The removal of a protecting group in process step (a) may also include the removal of R on the N-terminal group of a compound of formula I. For example, when R is benzyloxy carbonyl, this group may be removed by hydrogenation in the presence of a catalyst, e.g. Pd.

In accordance with process step (b) for example, for the production of a compound of formula I wherein X comprises a carboxy group, a compound of formula I wherein X comprises an esterified or amidated carboxy group may be hydrolysed. Such hydrolysis may be effected by treatment with an appropriate alkali or by acid hydrolysis, for example in the presence of trifluoroacetic acid.

Furthermore, in accordance with process step (b), for the production of a compound of formula I wherein X comprises an esterified or amidated carboxy group, a compound of formula I wherein X comprises a carboxy group or an esterified caboxy group may be (trans) esterified or amidated. Such ester formation or amidation may be carried out using any of the techniques known in the art, for example converting the carboxy group in a functional

reactive group, e.g. a corresponding carbonyl halide or anhydride, or using a compound of formula I wherein X is a radical of formula (h) in the lactone form, and reacting such group with the selected alcohol or amine.

In accordance with a further embodiment of process step (b), for the production of a compound of formula I wherein X is a radical of formula (g), a compound of formula I wherein X is a radical of formula (e_1) or (e_2) and A_5 is H may be reacted with a compound of formula II

$$H_2N\text{-}NH\text{-}CO\text{-}A_5 \qquad (II)$$

wherein A_5 is as defined above. This process may be carried out in analogy to the known techniques used for the preparation of semi carbazones.

A compound of formula I wherein X is a radical of formula (e_1) may also be converted in accordance with known techniques into a compound of formula I wherein X is a radical of formula (e_2) and vice versa.

Process step (c) may be carried out by the techniques known in the art of peptide chemistry. By peptide fragment comprising a radical of formula (e_1) to (j) is also meant the radical itself bearing a protecting group on the $-NR_6-$ moiety and an appropriate ending, e.g. A_5 or $CH_2\text{-}Z_a$, on the other end.

Process step (d) may conveniently be effected using a Dess-Martin reagent, e.g. in the presence of a base or a halogen-precipitating silver salt, or according to the Swern oxidation procedures.

Where desired, in these reactions, protecting groups may be used for functional groups which do not participate in the reaction.

These may be e.g. amino protecting groups, carboxy protecting groups, acetal groups etc. When the desired reaction is complete, the protecting groups may then be removed.

Each of the above processes may be carried out using starting materials in the form of one or other of the individual optical isomers or in the form of mixtures [relating to the asymetric carbons present in radicals of formulae (e_1) to (j) as X or in such a radical precursor]. Conveniently the starting materials are used as S- or R-enantiomers to produce a compound of formula I wherein the asymetric carbon in radicals of formulae (e_1) to (j) has the S or R configuration, respectively.

The starting materials used in process steps (d) or (e) may be prepared in analogy with process step (c).
Insofar as the production of the starting materials is not particularly described, the compounds are known or may be prepared analogously to methods known and practiced in the art.

The following examples are illustrative of the invention. All temperatures are in °C.

The following abbreviations are used:

THF = tetrahydrofuran
TFA = trifluoroacetic acid
MeOH = methanol
EtOAc = ethyl acetate
DCC = dicyclohexylcarbodiimide
HOBT = hydroxybenzotriazole
Z = benzyloxycarbonyl
r.t. = room temperature
Fmoc = 9-fluorenylmethoxycarbonyl
a = amorph

EXAMPLE 1: Z-Val-Met-Asp(OH)-H

Z-Val-Met-Aspartic aldehyde dimethyl acetal-β-tert.butyl ester (1.17 g) is taken up in CH_2Cl_2 (150 ml), TFA (15 ml) and water (1 ml) added and stirred for 2,5 hrs at r.t. The solvent is evaporated, toluene added twice and evaporated again. The crystalline residue is dissolved in water (60 ml) upon heating, decanted from some undissolved material and crystallized at 5 °, affording the first crop of the product: m.p. 117 °. All mother liquors are collected and chromatographed (SiO_2, acetone/hexane/-EtOAc 60/40/0.5) to give a second crop of product, crystallizing upon trituration with water.

EXAMPLE 2: Z-Val-Met-Asp(OH)-H Semicarbazone

Compound of Example 1 (0.27 g, 0.55 mmol) in MeOH (2 ml) is combined with semicarbazide, HCl (0.6 ml of 1 molar solution), then 5 drops of pyridine are added. The product crystallizes after 10 min. at r.t.: m.p. 233 - 235 °.

EXAMPLE 3: Z-Val-Met-Asp(OH)-H semicarbazonyl-Pro-Val-$N(CH_3)_2$

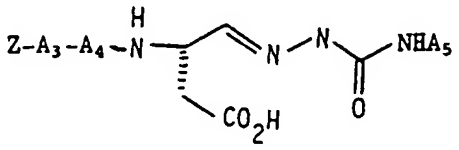
Compound of Example 1 (0.48 g, 1 mmol) is dissolved in a mixture of MeOH (2 ml) water (0.5 ml) and 3 drops of pyridine. N-(hydrazinocarbonyl)-Pro-Val-$N(CH_3)_2$ (0.3 g, 1 mmol) is dissolved in MeOH (1 ml), water (2 ml) and 3 drops of 2N HCl. Both solutions are combined and warmed for 2 min. at 40 - 50 °. The product crystallizes upon cooling: m.p. 118 - 120 °.

By repeating the procedure of Example 1, using the corresponding starting materials, following compounds may be prepared:

Example 4 Z-Val-Phe-Asp(OH)-H

Example 5 Z-Val-His-Asp(OH)-H

By repeating the procedure of Example 2 or 3 respectively, using the corresponding starting materials, following compounds of formula IA



(IA)

wherein A_3, A_4 and A_5 are as defined below, may be prepared.

Example	A_3	A_4	A_5	M.P.°C
6	Val	Phe	H	a
7	Val	His	H	150
8	Val	Phe	Pro-Val-$N(CH_3)_2$	126-129
9	Val	His	Pro-Val-$N(CH_3)_2$	
10	Ala-Tyr-Val	Phe	H	205

EXAMPLE 11: (3S)-3-(Z-valyl-phenylalanyl)-amino-5-(2,6-dimethyl benzoyloxy)-4-oxo pentanoic acid

(3S,4RS)-3-(Z-Val-His)amino-4-hydroxy-5-(2,6-dimethylbenzoyloxy) pentanoic acid tert.-butyl ester (0.6 mmol) is treated with Dess-Martin reagent (0.77 mmol) in CH_2Cl_2 (4 ml) for 45 min. and filtered. Aceton and 0.5 N NaOH are then added and aceton is evaporated. The residual crystalline material is washed with H_2O, acidified with 10 % tartaric acid and extracted with EtOAc. The combined EtOAc extracts affords after evaporation the tert. butyl ester of the product as yellow crystals. These are dissolved in CH_2Cl_2 (4 ml), TFA (4 ml) is added and the mixture is stirred at r.t. for 15 min. The mixture is evaporated to dryness and chromatographed (SiO_2, aceton/hexane/EtOAc 60/40/1) affording the product as slightly colored crystals.

EXAMPLE 12: (Z-valyl-alanyl)-(3R,4S)-3-amino-4-hydroxy-5-(2,6-dichlorbenzoyloxy)

(Z-valyl-alanyl)-3R,4S)-3-amino-4,5-dihydroxy pentanoic acid ethyl ester and 4-dimethylaminopyridine are dissolved in pyridine and 2,6-dichlorobenzoyl chloride is added dropwise. The reaction mixture is stirred overnight at room temperature, then ice and water are added and the mixture is extracted with AcOEt. The organic layer is washed with water, then with NaCl solution, dried over Na_2SO_4, filtered, evaporated, and the crude product is chromatographed to give the title compound.

The (3R,4RS) derivative of the title compound may be prepared as follows:

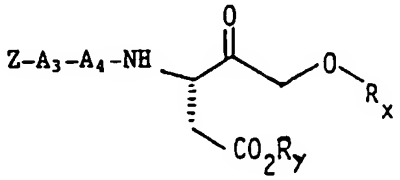
Z-Val-Ala-OH (0.26 mmol), DCC (53 mg, 0.26 mmol) and HOBT.H_2O (39 mg, 0.26 mmol) are dissolved in THF/DMF (2 ml/2 ml) and stirred for 5 min., before (3S,4RS)-3-amino-4-hydroxy-5(2,6-dimethylbenzoyloxy)-pentanoic acid tert.butyl ester (87 mg, 0.26 mmol) in THF (2 ml) is added. The reaction mixture is stirred overnight at r.t., evaporated and chromatographed (SiO_2, EtOAc/MeOH/NH_3 95/5/0.5) to afford the (3R,4RS) title compound.

EXAMPLE 13: (Z-valyl-alanyl)-(3R)-3-amino-4-oxo-5-(2,6-dichloro-benzoyloxy) pentanoic acid ethyl ester

Dimethylsulfoxide in CH_2Cl_2 is added dropwise to a solution of oxalyl chloride in CH_2Cl_2 at -50°C. After 15 min., a solution of compound of Example 12 in CH_2Cl_2 is added dropwise and the reaction mixture stirred for 1 hour at -40°C. Triethylamine is added and stirring is continued for 3 hours at room temperature. Water is then added and the reaction mixture extracted with CH_2Cl_2. The organic layer is washed with $NaHCO_3$ solution and

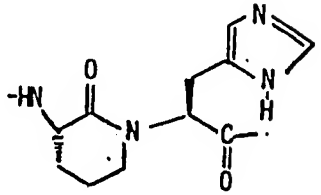
NaCl solution, dried over Na_2SO_4, filtered and evaporated. The residue is chromatographed yielding the title compound.

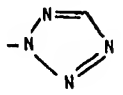
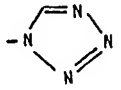
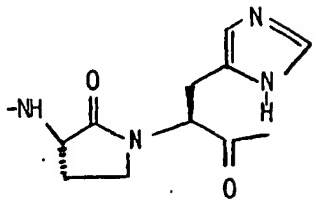
By repeating the procedure of Examples 1-3 and 11 to 13, using the corresponding starting materials, following compounds of formula IB



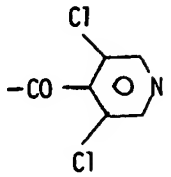
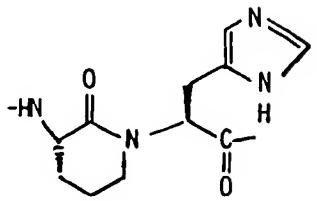
(IB)

wherein A_3, A_4, R_x and R_y are as defined below, may be prepared.

EXAMPLE	A_3	A_4	R_y	R_x	$[\alpha]_D^{20}$ or M.P.	c(MeOH) °C (2)
14	Val	Phe	H	2,6-diMe-benzoyl		
15	Val	His	H	2,6-diMe-benzoyl		
16	Val	His	H	p-NO_2-phenyl		
17	Val	His (1-trityl)	H	H	a (2)	
18	Val	His	H	2,6-diCl-benzoyl		
19	Val	His	H	2,6-diCF_3-benzoyl		
20	Val	His	H	2,6-diNO_2-benzoyl		
21	Val	His (2-CF_3)	H	2,6-diCF_3-benzoyl		
22	Val	Trp	H	2,6-diCF_3-benzoyl		
23	TriMe-silyl-Ala	His	H	2,6-diCl-benzoyl		
24			H	2,6-diCl-benzoyl		

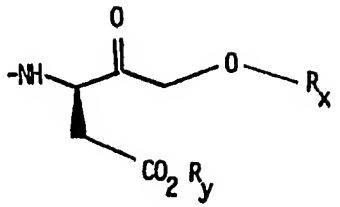
EXAMPLE	A_3	A_4	R_y	R_x	$[\alpha]_D^{20}$ or M.P.	c(MeOH) °C (2)
25	Ala-Tyr-Val	Phe	H	H	a (2)	
26	D-Val	His	H	2,6-diCl-benzoyl	-39.2°	1.21
27	Val	Arg	H	2,6-diCl-benzoyl		
28	Val	His	C_2H_5	2,6-diCl-benzoyl	-26.7°	1.02
29	Val	His	H		-18.0°	0.97
30	Val	His	H		-19.0°	0.96
31			H	2,6-diCl-benzoyl		

EXAMPLE	A_3	A_4	R_y	R_x	$[\alpha]_D^{20}$ or M.P.	c(MeOH) °C (2)
32	Val	Orn	H	2,6-diCl-benzoyl		
33	Val	Lys	H	2,6-diCl-benzoyl		
34	Val	Ala	H	2,6-diCl-benzoyl		
35	Val	His	H	Pivaloyl		
36	Val	His	i.C_4H_9	2,6-diCl-benzoyl	**	
37	Val	His	n.$C_{10}H_{21}$	2,6-diCl-benzoyl	**	
38	Val	His	benzyl	2,6-diCl-benzoyl	**	
39	Val	His	CH_2CH_2OH	2,6-diCl-benzoyl	**	
40	Val	Ala	C_2H_5	2,6-diCl-benzoyl	**	
41	Val	Leu	H	2,6-diCl-benzoyl		
42	Val	His	H	2,6-diF-benzoyl		
43	Val	His ($2\text{-}CF_3$)	H	2,6-diCl-benzoyl		
44	Val	His	C_2H_5	Pivaloyl	**	
45	Val	His	H	2,6-diCl-benzoyl	*	
46	Val	Lys ($N^{\varepsilon}\text{-}iC_3H_7$)	H	2,6-diCl-benzoyl		
47	Val	Lys ($N^{\varepsilon}\text{-}iC_3H_7$)	C_2H_5	2,6-diCl-benzoyl	**	
48	Val	Ala	H	2,6-diCl-benzoyl	**	
49	Val	Ala	t.C_4H_9	2,6-diCl-benzoyl	**	

EXAMPLE	A_3	A_4	R_y	R_x	$[\alpha]_D^{20}$ or M.P.	c(MeOH) °C (2)
50	Val	Ala	C_2H_5	2,6-diCl-benzoyl	*	
51	Val	Ala	C_2H_5		*	
52	TriMe-silyl-Ala	His	C_2H_5	2,6-diCl-benzoyl		
53			C_2H_5	2,6-diCl-benzoyl	**	
54	Val	Ala	i.C_3H_7		**	
55	TriMe-silyl-Ala	Ala	t.C_4H_9	2,6-diCl-benzoyl		
56	TriMe-silyl-Ala	Ala	H	2,6-diCl-benzoyl		

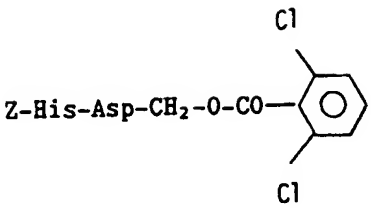
EXAMPLE	A_3	A_4	R_y	R_x	$[\alpha]_D^{20}$ or M.P.	c(MeOH) °C (2)
57	TriMe-silyl Ala	Ala	C_2H_5	2,6-diCl-benzoyl		

* the compounds have the following configuration in the aspartyl moiety:



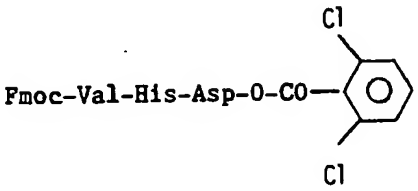
** the compounds have the R,S configuration in the aspartyl moiety.

EXAMPLE 58:

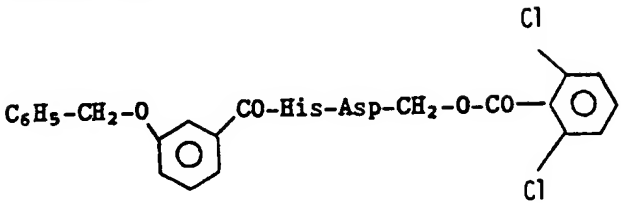


$[\alpha]_D^{20}$ = -20.3° c = 1 in MeOH

EXAMPLE 59:

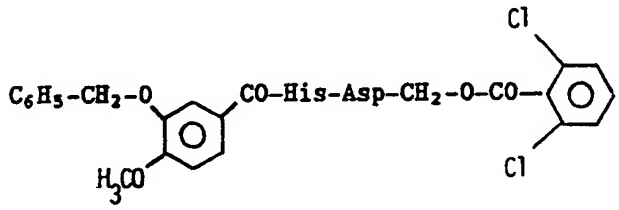


EXAMPLE 60:

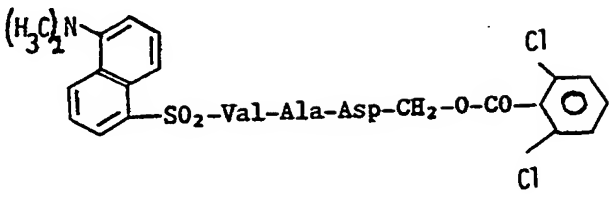


$[\alpha]_D^{20}$ = -21.5° c = 1.07 MeOH

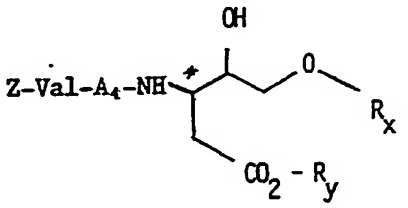
EXAMPLE 61:



EXAMPLE 62:

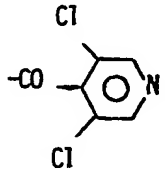


By repeating the procedure of Examples 1-3 and 12-13 and using the corresponding starting materials, following compounds of formula IC



IC

wherein A_3, R_x and R_y are as defined below, may be prepared

EXAMPLE	A_4	R_y	R_x	*
63	Ala	H	2,6-diCl-benzoyl	R,S
64	Ala	C_2H_5		R
65	Ala	t.C_4H_9	2,6-diCl-benzoyl	R,S
66	Ala	$-C_2H_4-OCH_3$	2,6-diCl-benzoyl	R,S

EXAMPLE 67: (3RS)-3-(Z-valyl-phenylalanyl)amino-4-oxo-6-(p-nitrophenyl)-transhex-5-enoic acid tert.butyl

(3RS)-3-(Z-Valyl-phenylalanyl)amino-4-oxo-pentanoic acid tert.butyl ester-5-diethylphosphonate (0.2 g, 0.29 mM) is dissolved in THF (15 ml) at 5°C. NaH (25 mg, 0.59 mM) is added and the mixture stirred for 10 min. p-Nitrobenzaldehyde (0.18 g, 1.1 mM) in THF (2 ml) is added at 0°C and the reaction mixture stirred for 45 min. at this temperature. The reaction mixture is poored on 5 % tartaric acid, extracted with ethyl acetate, the organic phases dried over Na_2SO_4, evaporated and the product purified by chromatography yielding the title compound.

EXAMPLE 68: Z-Val-Phe-Aspartic aldehyde dimethyl acetal-β-methyl ester

Z-Val-Phe-Asp(OH)-H (0.8 g) is dissolved in MeOH (25 ml) containing 8 % HCl, left at r.t. over night, evaporated to dryness and the residual crystals washed with ether, providing the title compound as white crystals. M.p.: 168 - 171 °.

Starting materials may be prepared as follows:

EXAMPLE 69: (3S)-3-(Fluorenylmethoxycarbonyl)amino-5-iodo-4-oxo butanoic acid tert.butyl ester

Triethylamine (1.3 ml, 9.6 mmol), followed be ethylchloroformiate (0.92 ml, 9.6 mmol) are added to (3S)-3-(fluorenylmethoxycarbonyl)amino-3-carboxy-butanoic acid tert.butyl ester (3.5 g, 8.5 mmol) in THF (60 ml) at - 10 °. After 10 min., a solution of diazomethane in ether is added slowly, and the reaction mixture stirred for 45 min. at 0 - 5 °. HCl (2N) in ether is added at 5 - 10 ° until gas evolution has ceased. The reaction mixture is evaporated to dryness, taken up in acetone (50 ml) NaI (4 g) added

and stirred for 1 hr at r.t. Ether (150 ml) is added, the reaction mixture filtered and evaporated. The residue is chromatographed (SiO_2, EtOAc/hexane), yielding the title compound as slightly yellow crystals.

EXAMPLE 70: (3S)-3-(Fluorenylmethoxycarbonyl)amino-5(2,6-dimethylbenzoyloxy)-4-oxo pentanoic acid tert.butyl ester

3S-3-(Fluorenylmethoxycarbonyl)amino-5-iodo-4-oxo-butanoic acid tert.butyl ester (1 g, 1.8 mmol), 2,6-dimethylbenzoic acid (0.5 g, 3.3 mmol) and AgOAc (0.6 g, 3.6 mmol) are dissolved in acetone (25 ml) and refluxed for 1 hr. After filtration and evaporation the crude product is chromatographed (SiO_2, ether/hexane 3/7) yielding the title compound.

EXAMPLE 71: (3S)-3-Fluorenylmethoxycarbonyl)amino-5(4-nitrophenoxy)-4-oxo pentanoic acid tert.butyl ester

3S-3-(Fluorenylmethoxycarbonyl)amino-5-iodo-4-oxo-butanoic acid tert.butyl ester (1 g, 1.8 mmol), p-nitrophenol (0.5 g, 3.7 mmol) and K_2CO_3 (0.38 g, 2.8 mmol) are refluxed in acetone (6 ml) for 30 min., CH_2Cl_2 added and the organic phase washed with 2N $NaHCO_3$. The combined organic phases are dried, evaporated and chromatographed (SiO_2, EtOAc/hexane 2/8), yielding the title compound as a yellow oil.

EXAMPLE 72: (Z-valyl-alanyl)-(3R,4S)-3-amino-4,5-dihydroxy pentanoic acid ethyl ester

a) Ethyl (3R,4S)-3-benzylamino-4,5-(isopropylidenedioxy) pentanoate (Y. Yamada, Tetrahedron Leters 1983, 24, 3009) and 10% Pd/C in ethanol are shaken at r.t. for 30 mn under H_2.

Filtration and evaporation of the reaction mixture give ethyl (3R,4S)-3-amino-4,5-(isopropylidenedioxy)pentanoate which is used without further purification.

b) Z-Val-Ala-OH is dissolved in THF, $HOBTH_2O$ and DCC are added at 5°C. After stirring for 20 min. at 5°C, diisopropylethylamine and ethyl (3R,4S)-3-amino-4,5-(isopropylidenedioxy)pentanoate in THF are added. Reaction mixture is stirred overnight at room temperature, filtered, evapored and chromatographed to afford (Z-valyl-alanyl)-(3R,4S)-3-amino-4,5-(isopropylidenedioxy)pentanoic acid ethyl ester.

c) Compound 74 b) is dissolved in $AcOH/H_2O$ (75/25) and stirred at 40°C for 4 hours. After evaporation, water is added and the mixture is extracted with AcOEt. The combined extracts are washed with water, $NaHCO_3$ solution, NaCl solution, dried over Na_2SO_4, filtered and evaporated, yielding the title compound.

The compounds of formula I their physiologically-hydrolysable and -acceptable esters and amides and their pharmaceutically acceptable salts (hereinafter referred to as compounds of the invention) exhibit pharmaceutical activity and are, therefore, useful as pharmaceuticals.

In particular, the compounds of the invention inhibit IL-1β secretion as indicated in the following in vitro test using THP-1 cells and in vivo test methods:

a) 900 µl THP-1 cells (0.5×10^6 cells) together with 100 U γ-interferon/0.9 ml RPMI 1640 medium (containing 2 mM L-glutamine and 5 % heat-inactivated foetal calf serum) are pipetted into 24 well culture plates. 100 µl of the compound to be tested are then added. After 3 hours at 37 ° C in 5 % CO_2/95 % air, 10 µl lipopolysaccharide 500 µg/ml is added and

the incubation continued for a further 40 hours. Appropriate controls (with and without stimulus, solvent) ar also included. The media are then removed and clarified by centrifugation at 1000 g for 10 min. 1.0 ml digitonin 0.01 % is added to the wells to lyse the cells which are loosened by scraping with a rubber policeman and left at 4 ° C for 10 min. Lactate dehydrogenase measurements are then performed immediately and the samples stored at - 20 ° C until the other determinations can be made. The assays are: IL-1β (medium and lysate), IL-6 (medium), TNF-α (medium), PGE2 (medium and lysate), lactate dehydrogenase (LDH) and DNA (lysates). IL-1β, IL-6 and TNF-α assays are determined using commercially available ELISA kits (Cistron), PGE_2 is measured using a standard RIA and DNA fluorimetrically using DAPI.

In this test, the compounds of the invention selectively inhibit IL-1β release in concentrations from about 0.01 to 100 µM. In contrast IL-6, TNF-α, PGE2 and DNA levels remain substantially unaffected, and the compounds are non-toxic, since LDH release is unchanged. It has for example been determined that compounds of examples 40 and 50 have an IC_{50} value (concentration of compound which inhibits to 50% the release of IL-1β) of 1 and 0.1 µM respectively.

b) LPS-Fever

A LPS-suspension (Sigma, No. L-5886; 100µg/5ml glucose solution/kg s.c.) is injected in male Tuttlingen SD rats (150-160g). 2 hours later the body temperature is measured using a thermistor rectal probe connected to an ELLAB telethermometer. After 4 hours the test compound is administered p.o. 2 hours later (6 hrs after LPS administration) the temperature is measured again. The temperature increment shown by the untreated controls is taken as 100% and that in

the treated group is expressed as a percentage of this value. The ED_{50} is the dose causing a 50% inhibition of the temperature increase determined in the control rats. In this test compounds of the invention inhibit the LPS-induced temperature increase when administered at a dosage in the range of from 0.001 to 0.1 mg/kg p.o. It has for example been determined that compounds of examples 40 and 50 have each an ED_{50} value of 0.01 mg/kg p.o. and compound of example 51 an ED_{50} value of 0.05 mg/kg p.o.

c) Carrageenan-Induced Paw Edema in the Rat

50FA male rats, 150-170g body weight, are used for each group. The test compound is administered orally as a suspension in physiological saline/0.5% tragacanth 1 hour prior to the carrageenan injection. Carrageenan (0.1ml of a 1% suspension in physiological saline) is given by subplantar injection into one hind paw. The swelling of the paw is measured by means of an antiphlogometer according to Kemper & Amelm. A control reading is taken immediately after the injection, and the swelling is measured after 3 and 5 hrs. The mean value of the 3- and 5-hour reading is taken after deduction of the control reading, the values obtained from the treated animals are expressed as a percentage of the value obtained from non-treated controls. The ED_{50} is the dose causing a 50% inhibition of the carrageenan-induced swelling after 3 hrs. In this test method compounds of the invention inhibit significantly the carrageenen-induced swelling when administered p.o. at a dosage in the range of from 0.02 to 5 mg/kg. It has for example been determined that compound of example 40 and 50 have an ED_{50} value of 0.2 and 1 mg/kg p.o. respectively.

Compounds of the invention are therefore useful for the treatment of disorders with an aetiology associated with or comprising excessive IL-1β release, e.g. in a wide variety of inflammatory states and diseases, for example tissus calcium depletion, degenerative processes in bone and cartilage, e.g. rheumatoid arthritis and osteoarthritis, inflammatory bowel disease, irritable bowel disease, septic shock, psoriasis, asthma, adult respiratory distress syndrome, diabetes type I, osteoporosis of various genesis including e.g. climacteric or post-menopausal osteoporosis as well as osteoporosis consequential to old age, immobilization or trauma, arteriosclerosis and Alzheimer disease.

For the above uses the required dosage will of course vary depending on the mode of administration, the particular condition to be treated and the effect desired. In general however, satisfactory results are achieved at daily dosage rates of from about 0.001 to about 100 mg/kg, preferably 0.001 to about 10 mg/kg animal body weight. Suitable daily dosage rates for larger mammals, for example humans, are of the order of from about 0.1 mg to about 1 g/day, conveniently administered once, in divided dosages 2 to 4 x/day, or in sustained release form.

Compounds of Examples 50 and 51 are preferred.

In accordance with the foregoing the present invention also provides:

a) A method for the treatment of disorders with an aetiology associated with or comprising excessive IL-1β release, e.g. as indicated above in a subject in need thereof, which method comprises administering to said subject an effective amount of a compound of formula I, a physiologically-hydrolysable and -acceptable ester or amide thereof, or a pharmaceutically acceptable salt thereof;

b) A compound of formula I, a physiologically-hydrolysable and -acceptable ester or amide thereof or a pharmaceutically acceptable salt thereof, for use as a pharmaceutical, for example for use as an agent, e.g. in the method as disclosed above.

The compounds of the invention may be administered by any conventional route, in particular nasally, enterally, preferably orally, e.g. in the form of tablets or capsules, or parenterally e.g. in the form of injectable solutions or suspensions or in a suppository form. Unit dosage forms contain, for example from about 25 µg to 500 mg of a compound of the invention.

The compounds of the invention may be administered in free form or in pharmaceutically acceptable salt form. Such salts may be prepared in conventional manner and exhibit the same order of activity as the free compounds.

Furthermore the present invention also provides:

c) A pharmaceutical composition comprising a compound of formula I, a physiologically-hydrolysable and -acceptable ester or amide thereof or a pharmaceutically acceptable salt thereof, as hereinbefore defined, together with a pharmaceutically acceptable diluent or carrier therefor. Such compositions may be manufactured in conventional manner. They may comprise up to 99.9% by weight of active ingredient.

CLAIM

1. A compound of formula I

$$R-[A_1-A_2]_n-A_3-A_4-X-A_5 \qquad (I)$$

wherein

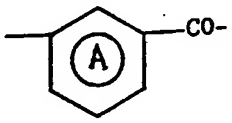
R is hydrogen, an amino protecting group or optionally ring substituted benzyloxy

n is 0 or 1,

A_1 is Val, Leu, Ala, Ile or trimethylsilyl-Ala

A_2 is Phe or Tyr,

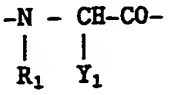
A_3 is a direct bond, Val, Leu, Ala, Ile, trimethylsilyl-Ala or a divalent radical of formula (a)



(a)

wherein ring A is optionally substituted by hydroxy or C_{1-4}alkoxy,

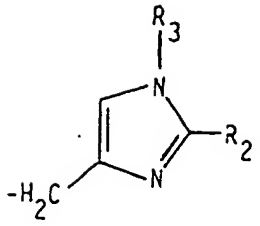
A_4 is a direct bond or a divalent radical of formula (b)



(b)

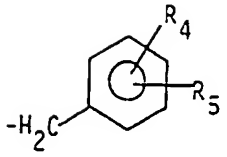
wherein R_1 is hydrogen or C_{1-4}alkyl, and

Y_1 is the residue attaching to the α-carbon atom of an α-amino acid and optionally protected, $-CH_2-CH_2-N(C_{1-4}alkyl)_2$, imidazol-2-yl-methyl, benzimidazol-2-yl-methyl, 1H-1,2,4-triazol-3-yl-methyl, pyrazol-3-yl-methyl, indazol-3-yl-methyl or a radical of formula (c) or (d)



(c)

or



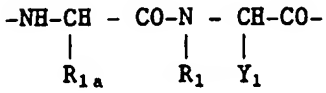
(d)

wherein

each of R_2 and R_3, independently, is hydrogen, halogen, C_{1-4}alkyl, CF_3 or trityl, at most one of R_2 and R_3 being H, and

each of R_4 and R_5 independently is hydrogen, C_{1-4}alkyl, hydroxy, C_{1-4}alkoxy, CF_3, phenyl or halogen, at most one of R_4 and R_5 being H,

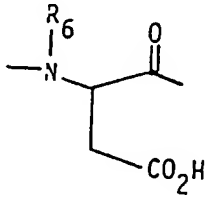
or A_3 and A_4 form together a radical of formula (aa)



(aa)

wherein Y_1 is as defined above and R_1 and R_{1a} form together $-(CH_2)_m-$ wherein m is 2, 3, 4 or 5, and

i) X is a divalent radical of formula (e_1)



(e_1)

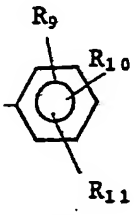
wherein R_6 is H or C_{1-4}alkyl,

and A_5 is hydrogen; CF_3; a radical $-Z_1-Z_2-Y_2$ wherein each of Z_1 and Z_2 independently is a direct bond or an α-amino acid residue and Y_2 is NH_2, C_{1-4}alkylamino, di-(C_{1-4}-alkyl)amino or a heterocyclic radical attached by a

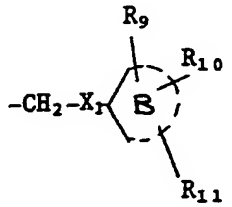
nitrogen to Z_2; a radical $-CH_2-X_1-Y_3$ wherein X_1 is O or S and Y_3 is heteroaryl; a radical $-CH_2-Y_3$; or a radical of formulae (k) to (m)

(k) (l)

or



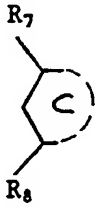
(m)



(o)

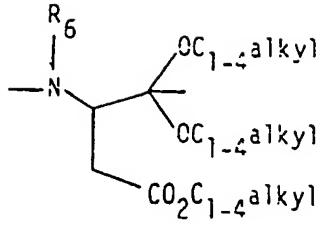
wherein

Y_4 is tri-(C_{1-4}alkyl)methyl or a residue



ring B is pyridyl,
ring C is phenyl or pyridyl,
each of R_7 and R_8 independently is C_{1-4}alkyl, C_{1-4}alkoxy, CF_3, halogen, nitro or cyano, and
each of R_9, R_{10} and R_{11} independently is nitro, cyano, CF_3, carbamoyl, CO_2R_{12}, $-CH=CH-CN$ or $-CH=CHCO_2R_{12}$ wherein R_{12} is C_{1-6}alkyl,

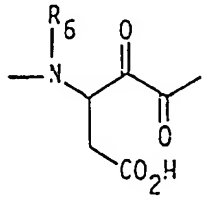
X being also a divalent radical of formula (e_2)



(e_2)

when A_5 is H, or

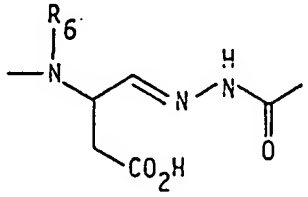
ii) X is a divalent radical of formula (f),



(f)

and A_5 is $-Z_1-Z_2-Y_2$ or a radical of formulae (k) to (o) as defined above, or OR_{13} or $NR_{14}R_{15}$ wherein R_{13} is C_{1-12}alkyl optionally substituted by OH or interrupted by O and each of R_{14} and R_{15} is independently hydrogen, C_{1-12}alkyl, C_{5-7}cycloalkyl or benzyl, or

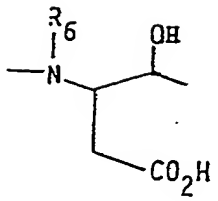
iii)X is a divalent radical of formula (g)



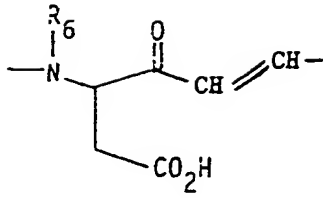
(g)

and A_5 is $-Z_1-Z_2-Y_2$ as defined above, or

iv) X is a divalent radical of formula (h) or (j)



(h)



(j)

and A_5 is a radical of formulae (k) to (o), $-CH_2-Y_3$ or $-CH_2-X_1-Y_3$ as defined above,

with the provisos that
only one of A_3 and A_4 can be a direct bond when n is 0, and each of A_3 and A_4 is other than a direct bond when n is 1.

and the physiologically-hydrolysable and -acceptable esters or amides thereof,

in free form, in salt form or in the form of complexes.

2. A compound of formula I according to claim 1, wherein n is 0, A_3 is a direct bond, Val, Leu, Ala, Ile or trimethylsilyl-Ala, A_4 is as defined in claim 1, or A_3 and A_4 form together a radical of formula (aa) as defined in claim 1, and
   i) X is a radical of formula (e_1) or (e_2) as defined in claim 1 and A_5 is H, or
   ii) X is a radical of formula (e_1) or (f) and A_5 is $-Z_1-Z_2-Y_2$ as defined in claim 1 or a radical of formula (k), (l) or (m) as defined in claim 1 wherein X_1 is O,
   iii) X is a radical of formula (g) and A_5 is $-Z_1-Z_2-Y_2$ as defined in claim 1, or
   iv) X is a radical of formula (j) and A_5 is a radical of formula (k), (l) or (m) as defined in claim 1 wherein X_1 is O.

and the physiologically-hydrolysable and -acceptable esters or amides thereof,
in free form, in salt form or in the form of complexes.

3. A compound of formula I according to claim 1 wherein n is 0 and each of A_3 and A_4 are other than a direct bond, and the physiologically-hydrolysable and -acceptable esters or amides thereof, in free form, in salt form or in the form of complexes.

4. A compound of formula I according to claim 1 or 3 wherein Y_1 is the residue attaching to the α-carbon atom of an α-amino acid selected from Ala, Leu, His, Phe, Met, Trp, trimethylsilyl-Ala and optionally side chain protected Arg, Orn and Lys, or a radical of formula (c), and the physiologically-hydrolysable and -acceptable esters or amides thereof, in free form, in salt form or in the form of complexes.

5. A compound of formula I according to claim 1, 3 or 4 wherein X is a radical of formula (e_1), (h) or (j), A_5 being other than H when X is (e_1), and the physiologically-hydrolysable and -acceptable esters or amides thereof, in free form, in salt form or in the form of complexes.

6. (Benzyloxycarbonyl-valyl-alanyl)-3R-3-amino-4-oxo-5-(2,6-dichlorobenzoyloxy) pentanoic acid ethyl ester, (benzyloxycarbonyl-valyl-alanyl)-3R,S-3-amino-4-oxo-5-(2,6-dichloro benzoyloxy) pentanoic acid ethyl ester, (benzyloxycarbonyl-valyl-alanyl)-3R-3-amino-4-oxo-5-(2,6-dichloro-pyridyl-4-carbonyloxy) pentanoic acid ethyl ester, (benzyloxycarbonyl-valyl-alanyl)-3R,S-3-amino-4-oxo-5-(2,6-dichlorobenzoyl oxy) pentanoic acid, (benzyloxycarbonyl-valyl-alanyl)-3R,S-3-amino-4-oxo-5-(2,6-dichlorobenzoyloxy) pentanoic acid isopropyl ester, (benzyloxycarbonyl-valyl-alanyl)-3R,S-3-

amino-4-oxo-5-(2,6-dichlorobenzoyloxy) pentanoic acid t.-butyl ester, and (benzyloxycarbonyl-valyl-alanyl)-3S--3-amino-4-oxo-5-(2,6-dichlorobenzoyloxy) pentanoic acid in free form, in salt form or in the form of complexes.

7. A process for producing a compound of formula I and the physiologically-hydrolysable and -acceptable esters and amides thereof, as defined in claim 1, which process comprises

a) removing at least one protecting group from a compound of formula I in protected form or adding a protecting group R as defined in claim 1 at the N-terminal group of a compound of formula I; or

b) converting one compound of formula I into another compound of formula I; or

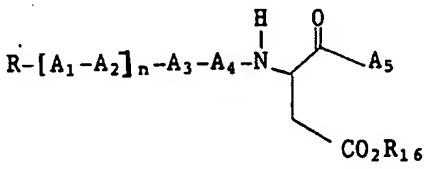
c) coupling together by an amide bond two peptide fragments, each of which contains at least one amino acid in protected or unprotected form and one peptide fragment containing a radical of formula (e_1) to (j) as defined in claim 1, the peptide fragments being such that a protected or unprotected peptide having the sequence according to formula I above is obtained and, if necessary, removing the protecting group or groups from a compound of formula I in protected form; or

d) for the production of a compound of formula I wherein X is a radical of formula (e_1) or (h) and A_5 is a radical of formula (k), (l) or (o) or $-CH_2-X_1-Y_3$ as defined in claim 1, reacting a compound of formula III

$$R-[A_1-A_2]_n-A_3-A_4-X'-CH_2-Z_a \qquad (III)$$

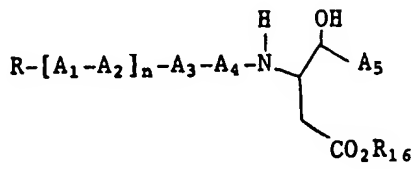
wherein R, A_1 to A_4 and n are as defined in claim 1, X' is a radical of formula (e_1) or (h) as defined in claim 1, and Z_a is a leaving group,
with a corresponding phenol, thiophenol or HX_1-pyridine or an acid of formula HX_1-CO-Y_4 or a functional derivative thereof or HX_1-Y_3 wherein X_1, Y_3 and Y_4 are as defined in claim 1; or

e) for the production of a compound of formula I



(I)

wherein R, A_1 to A_5 and n are as defined in claim 1 and R_{16} is a C_{1-12} aliphatic or alicyclic residue, oxidizing a compound of formula V



(V)

wherein R, A_1 to A_5, n and R_{16} are as defined above,

and recovering a compound of formula I or a physiologically-hydrolysable and -acceptable ester or amide thereof thus obtained in free or salt form or in the form of a complex.

8. A compound according to any one of claims 1 to 6 for use as a pharmaceutical.

9. A pharmaceutical composition comprising a compound of formula

I as defined in claim 1 or a physiologically-hydrolysable and -acceptable ester or amide thereof, in free form or in physiologically acceptable salt form, together with a pharmaceutically acceptable diluent or carrier therefor.

10. A method for the treatment of disorders with an aetiology associated with or comprising excessive IL-1β release, in a subject in need thereof, which method comprises administering to said subject an effective amount of a compound of formula I as defined in claim 1, a physiologically-hydrolysable and -acceptable ester or amide thereof, or a pharmaceutically acceptable salt thereof.

INTERNATIONAL SEARCH REPORT

International Application No PCT/EP 92/02472

I. CLASSIFICATION OF SUBJECT MATTER (if several classification symbols apply, indicate all)[6]

According to International Patent Classification (IPC) or to both National Classification and IPC

Int.Cl. 5 C07K5/04; C07K5/06; A61K37/02

II. FIELDS SEARCHED

Minimum Documentation Searched[7]

Classification System	Classification Symbols
Int.Cl. 5	C07K

Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched[8]

III. DOCUMENTS CONSIDERED TO BE RELEVANT[9]

Category °	Citation of Document,[11] with indication, where appropriate, of the relevant passages[12]	Relevant to Claim No.[13]
X	WO,A,9 115 577 (IMMUNEX CORPORATION) 17 October 1991 * See page 5, lines 5-26 * * See page 8, line 6 - page 9, line 37 * ---	1-5,7-10
X	VOELTER ET AL 'Tripeptides and fragments (Volume 3)' 1983 , GEORG THIEME VERLAG , STUTTGART, GERMANY * See page 180 * ---	1-2,4-5, 7
X	PETTIT ET AL 'Synthetic peptides (Volume 1)' 1970 , VAN NOSTRAND REINHOOLD COMPANY , NEW YORK, USA * See pages 205 and 211 * --- -/--	1-2,4-5

° Special categories of cited documents :[10]

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

IV. CERTIFICATION

Date of the Actual Completion of the International Search	Date of Mailing of this International Search Report
11 JANUARY 1993	29. 01. 93
International Searching Authority	**Signature of Authorized Officer**
EUROPEAN PATENT OFFICE	KORSNER S.E.

Form PCT/ISA/210 (second sheet) (January 1985)

III. DOCUMENTS CONSIDERED TO BE RELEVANT (CONTINUED FROM THE SECOND SHEET)

Category °	Citation of Document, with indication, where appropriate, of the relevant passages	Relevant to Claim No.
A	EDITORS: SMITH ET AL // AUTHORS: WEIDNER ET AL 'Proceedings 12th American Peptide Symposium, June 1991 // Comparison of peptide and protein substrates for interleukin-1beta convertase' , ESCOM , LEIDEN, HOLLAND * See pages 891-892 *	1-10
A	JOURNAL OF BIOLOGICAL CHEMISTRY vol. 265, no. 24, August 1990, BALTIMORE, USA pages 14526 - 14528 SLEATH ET AL 'Substrate specificity of the protease that processes human Interleukin-1beta' * See Discussion, pages 14527-8 *	1-10

INTERNATIONAL SEARCH REPORT

Int tional application No.
PCT/EP 92/02472

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:
Remark: Although claim 10 is directed to a method of treatment of the humen body, the search has been carried out and based on the alleged effects of the compounds.

2. [ ] Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful internaional search can be carried out, specifically:

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. [ ] As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. [ ] As all searchable claims could be searches without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest.

[ ] No protest accompanied the payment of additional search fees.

ANNEX TO THE INTERNATIONAL SEARCH REPORT ON INTERNATIONAL PATENT APPLICATION NO. EP 9202472 SA 66224

This annex lists the patent family members relating to the patent documents cited in the above-mentioned international search report. The members are as contained in the European Patent Office EDP file on 11/01/93
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Patent document cited in search report	Publication date	Patent family member(s)	Publication date
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